Quantifying *in situ* Zooplankton Movement and Trophic Impacts on Thin Layers in East Sound, Washington

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LONG-TERM GOALS

Our long-term goals are to quantify and predict how rapidly and effectively various functional groups of zooplankton aggregate to and exploit localized, transient resource concentrations, such as those represented by thin layers. We are particularly interested in the consequences of transient resource concentrations for the dynamics of planktonic communities over large space and time scales.

OBJECTIVES

The objectives of this study are:

(1) To quantify the degree to which micro-planktonic predators recruit to and exploit thin layers of phytoplankton resources in thin layers.

(2) To document the effects of high resource levels within thin layers on micro-predator reproduction and dispersal, and the trophic impacts of these predators on phytoplankton and other biotic components of thin layers.

(3) To synthesize these observations into a predictive modeling framework that translates from observations of individual cells at small space and time scales into an understanding of biological-physical interactions on the much larger space and time scales of oceanographic features such as thin layers.

APPROACH

Our approach has been to develop and deploy two novel sampling methods intended to quantify *in situ* micro-plankton dynamics.

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The first of these is a Vertical Structure Sampler (VSS) that encloses and retrieves for shipboard study a 2 m section of water column with intact vertical structure (Figure 1). The VSS is targeted at specific water column features identified by CTD profiles. The VSS is equipped with two replicate 20 l sampling cylinders, each having a smooth interior and a knife-edged cowling to minimize water disturbance during descent. Gate valves are remotely triggered at depth, whereupon the VSS is brought to the surface and suspended within an enclosure on deck that isolates it from ship roll and heave, and enables control of the light and temperature environment during subsequent incubation. The VSS is equipped with 20 ports at 10 cm vertical intervals, through which water samples can be extracted and inserted. The ports are provided with O-ringed pistons to exclude water when the port is not in use, to prevent contamination of samples by settling and colonization. An additional instrument, the Multisampler, resolves spatial structure by capturing water samples in an array of ten 200 ml sampling bottles that can be positioned anywhere along a lightweight, 2 m long backbone (Figure 1). The Multisampler can be deployed alongside the VSS to ground truth vertical microstructure, or can be deployed horizontally to quantify fine-scale horizontal variability in thin layer composition.

The second sampling method used in our study has been a field-deployable, side-by-side combination of our optical plankton tracking system with a FlowCAM imaging flow cytometer. The strength of the combination is that the tracking system quantifies swimming behaviors of protists in natural seawater samples with large numbers of motile and non-motile particles, while the FlowCAM simultaneously provides rapid identification of dominant taxonomic groups present in the same samples. The tracking system can be used on deck for analysis of VSS samples, or deployed with underwater housings to image *in situ* plankton behaviors. Trajectories obtained from these observations serve to parameterize spatially-explicit models of fine-scale plankton dynamics developed in earlier phases of this research.

WORK COMPLETED

In FY 2006, we conducted four cruises on R/V Barnes and R/V Centennial to East Sound (October 18-21, 2005, and May 2-5, June 22-24, and August 30-31, 2006). In addition, we participated in the LOC2006 cruise on R/V Thompson to Monterey Bay (July 8-30, 2006). Cumulatively, during these cruises we carried out 8 deployments of the VSS, together with associated FlowCAM analysis and preparation of fixed samples for taxonomic analysis of plankton abundance and distribution. We also captured video sequences for movement analysis on 70 water samples from East Sound and Monterey Bay.

RESULTS

The focus of our initial (October, 2005) cruise was to establish coordinated deployment and sampling protocols for the VSS, Multisampler and FlowCAM. These initial deployments also served to test the capability of the VSS to capture sections of water column and return them to the surface without disrupting their fine-scale vertical structure. Though we did not observe thin layers on this cruise, FlowCAM analysis of the VSS samples and subsequent analysis of preserved plankton samples support indications from laboratory tests that the VSS does return captured water samples with vertical structure substantially intact (Figure 2).

The focus of the May, June and August 2006 cruises to East Sound was to link observations of thin layers to behavioral analysis of protists resident above, within, and below these features. Analysis of our samples is at a very early stage. We present some initial results here to indicate the direction of our on-going research.

On May 3, 2006, we conducted CTD casts at East Sound sampling station #14 (122.902°W, 48.685°N), at 0806h, 1202h and 1537h (Figure 3). The profiles from these casts show the formation between morning and mid-afternoon of a plankton layer between 10 and 30 cm thick at 9.5 m depth, representing a greater than three-fold increase in chlorophyll a fluorescence over background. This layer is interesting because it is associated with only weak thermal and salinity stratification. The apparent weakness of physical structuring mechanisms suggests that behavioral mechanisms such as active swimming or buoyancy regulation may have played a role in formation of this thin layer.

Video analysis of East Sound water samples obtained the next day (May 4, 2006) show large numbers of diatom chains and smaller passive particles, that create a heavily occluded environment for motile ciliates and dinoflagellates (Figure 4, upper left image). To assess the potential role of active swimming and buoyancy regulation, we are presently analyzing trajectories of individual cells and diatom chains. We are not yet able to make statistical statements about swimming characteristics of the motile protists in our video samples. However, our early results demonstrate that we can track the large numbers of passive and active cells in our samples (which amount to hundreds in each video frame; Figure 4, bottom image). Furthermore, we can capture trajectories of active swimmers and distinguish their movements from those of surrounding passively sinking cells. An example is in the upper right image of Figure 4, which represents the trajectory of a large ciliate as it swam among diatoms and other particles and exhibited a number of distinctive behaviors, including helical swimming and "hopping".

An animation of this trajectory is available at:

http://peduncleii.ocean.washington.edu/~dg/ciliate3e.mov

Contemporaneous FlowCAM analysis of this water sample enabled us to identify the diatom chains in the video images as consisting predominantly of *Guinardia delicatula* and *Leptocylindrus danicus* (Figure 5). The dominant large ciliate, and the organism that is almost executed the long trajectory in Figure 4, is *Laboea strobila*. Other motile taxa present in large numbers were a number of other ciliates and dinoflagellates (Figure 5). As analysis of video and FlowCAM data and counting of fixed samples proceeds, we expect to be able to make inferences about how swimming and buoyancy regulation behaviors may have contributed to formation of thin layers such as the one we observed on May 3, 2006 in East Sound.

IMPACT/APPLICATIONS

This research has developed a significant new capability to quantify distribution patterns of motile and non-motile plankton taxa in the vicinity of thin layers and other fine-scale water column features. By enabling capture and ship-board study of thin layer sections, the Vertical Structure Sampler provides the opportunity to investigate ambient water column conditions, to analyze the time course of thin

layer dynamics in mesocosm-type culturing experiments, and to understand the consequences of manipulations of specific water strata. In combination with behavioral tracking and FlowCAM identification of plankton community composition, the methods developed in this research provide new and more direct techniques to assess relationships between population distributions and *in situ* behavioral characteristics across a range of plankton taxa.

RELATED PROJECTS

This research contributed to collaborative field work with T. J. Cowles (N000140410277) and other LOCO-funded ONR investigators, and to collaborative modeling work with P. S. J. Franks (N000140310391, N000140610304).

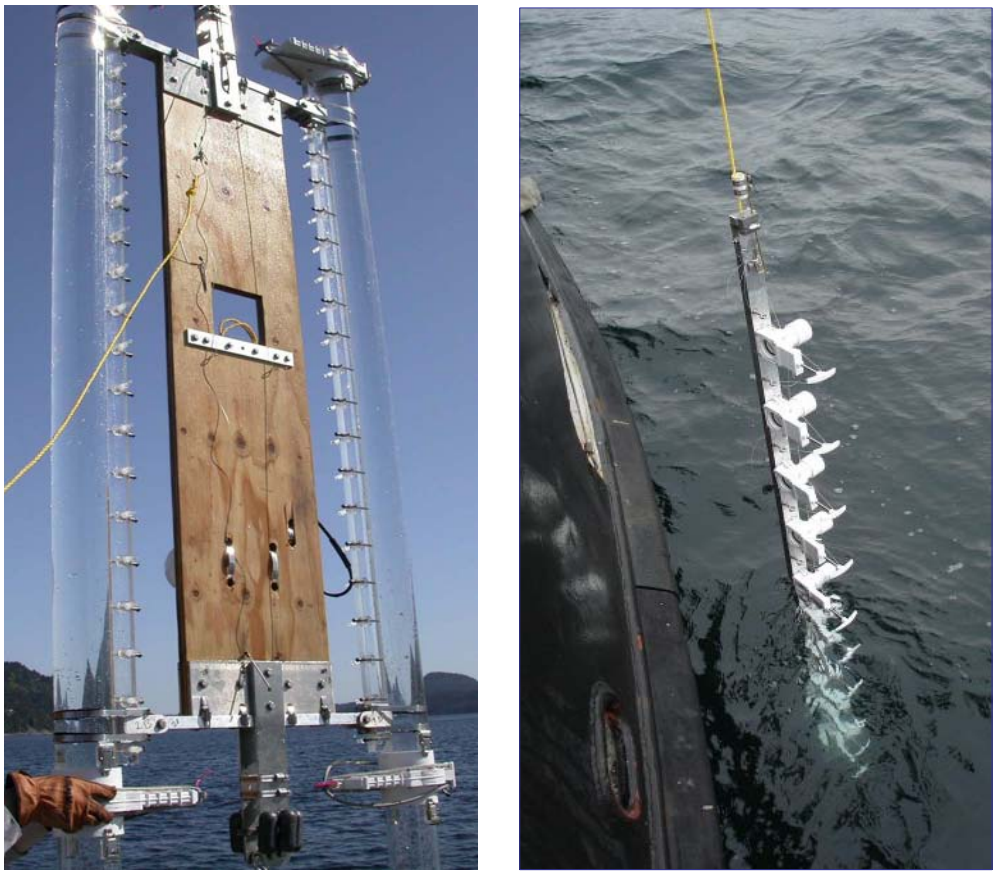

Figure 1. The Vertical Structure Sampler (VSS, left) and the Multisampler (right) being deployed from R/V Barnes in East Sound, WA (October, 2005.

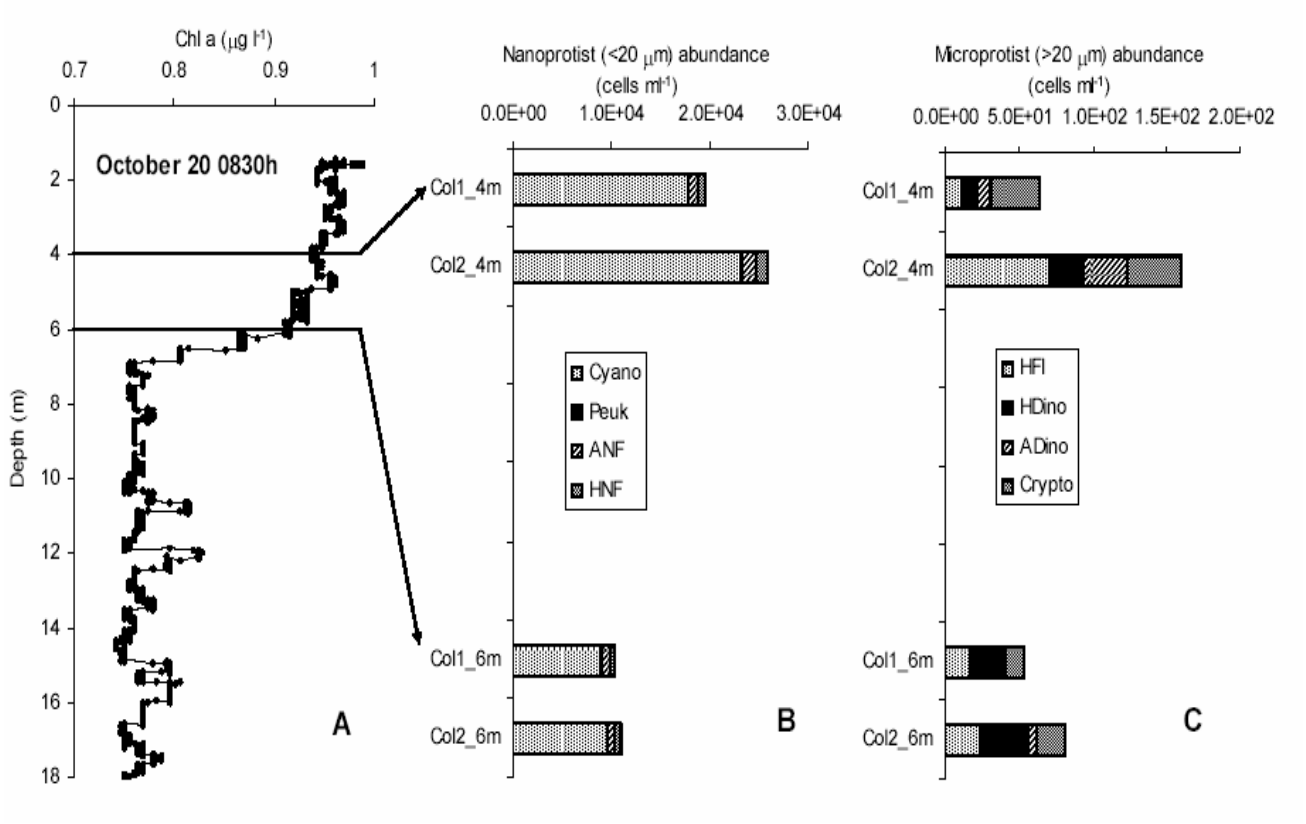

Figure 2. Left: CTD profile from East Sound on October 20, 2005 at 08:30, showing vertical structure of the chlorophyll signature. Horizontal lines at 4 m and 6 m represent the top and bottom positions of the VSS deployment into this distribution. Middle and right: Taxonomic categories of nano- and microprotists captured in the VSS showing intact vertical structure. Symbols: Cyanobacteria (Cyano), Non-motile Picoeukaryotes (Peuk), Autotrophic nanoflagellates (ANF), Heterotrophic nanoflagelates (HNF), Heterotrophic Microflagellates (HFl), Heterotrophic dinoflagellates (HDino), Autotrophic dinoflagellates (ADino), and Cryptophytes (Crypto).

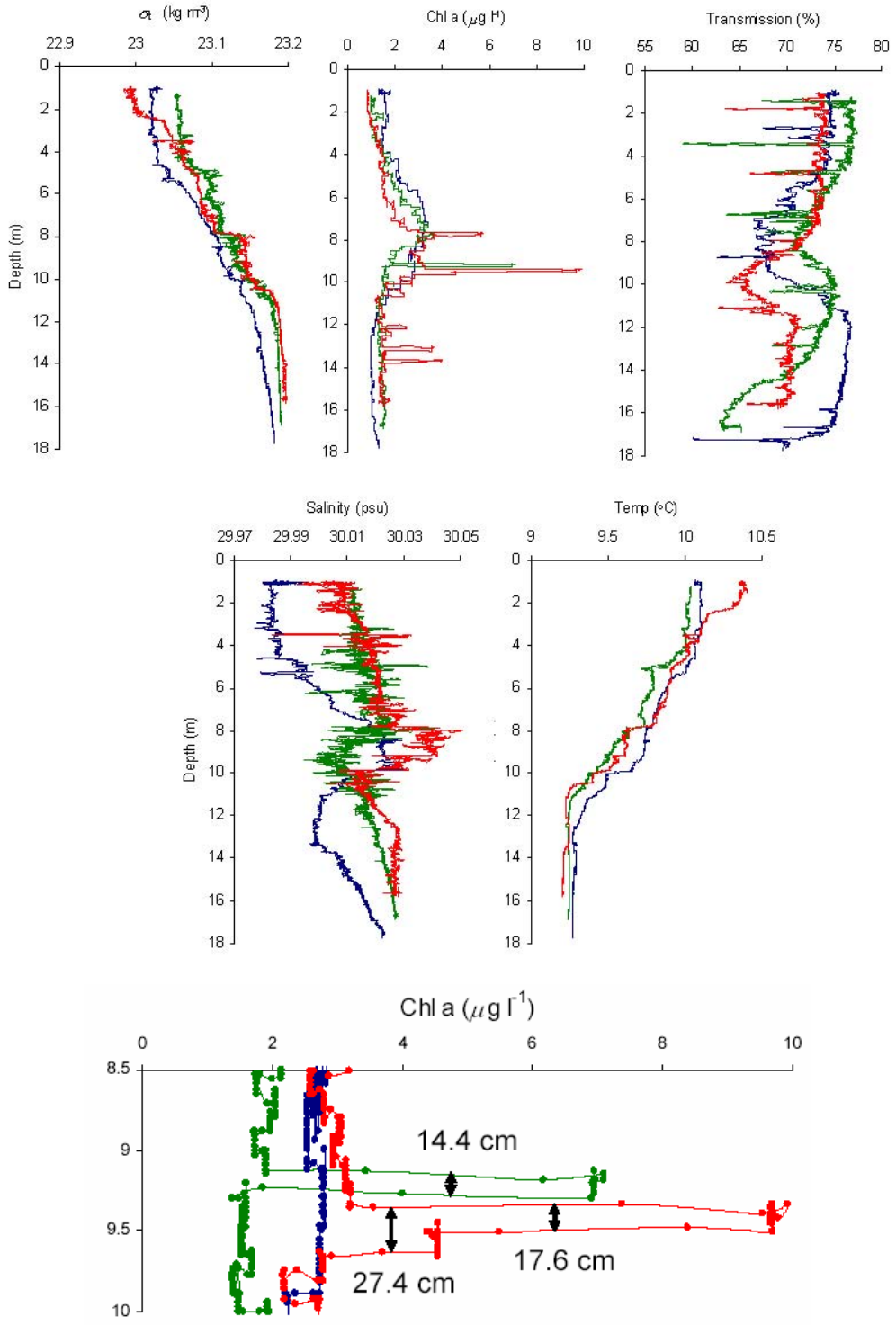

Figure 3. Top and middle: Profiles showing the formation of a biological thin layer in the presence of very weak thermal stratification in East Sound on May 3, 2006. Vertical profiles of σ_t, chlorophyll a ($\mu g\ l^{-1}$) derived from calibrated in vivo fluorescence, transmissivity (%), salinity (ppt), and temperature (°C) are shown for East Sound station 14 (122.902°W, 48.685°N), sampled at 0806h (----), 1202h (----) and 1537h (----). Bottom: Expanded view of the development of the chlorophyll peak at 9.5 m.

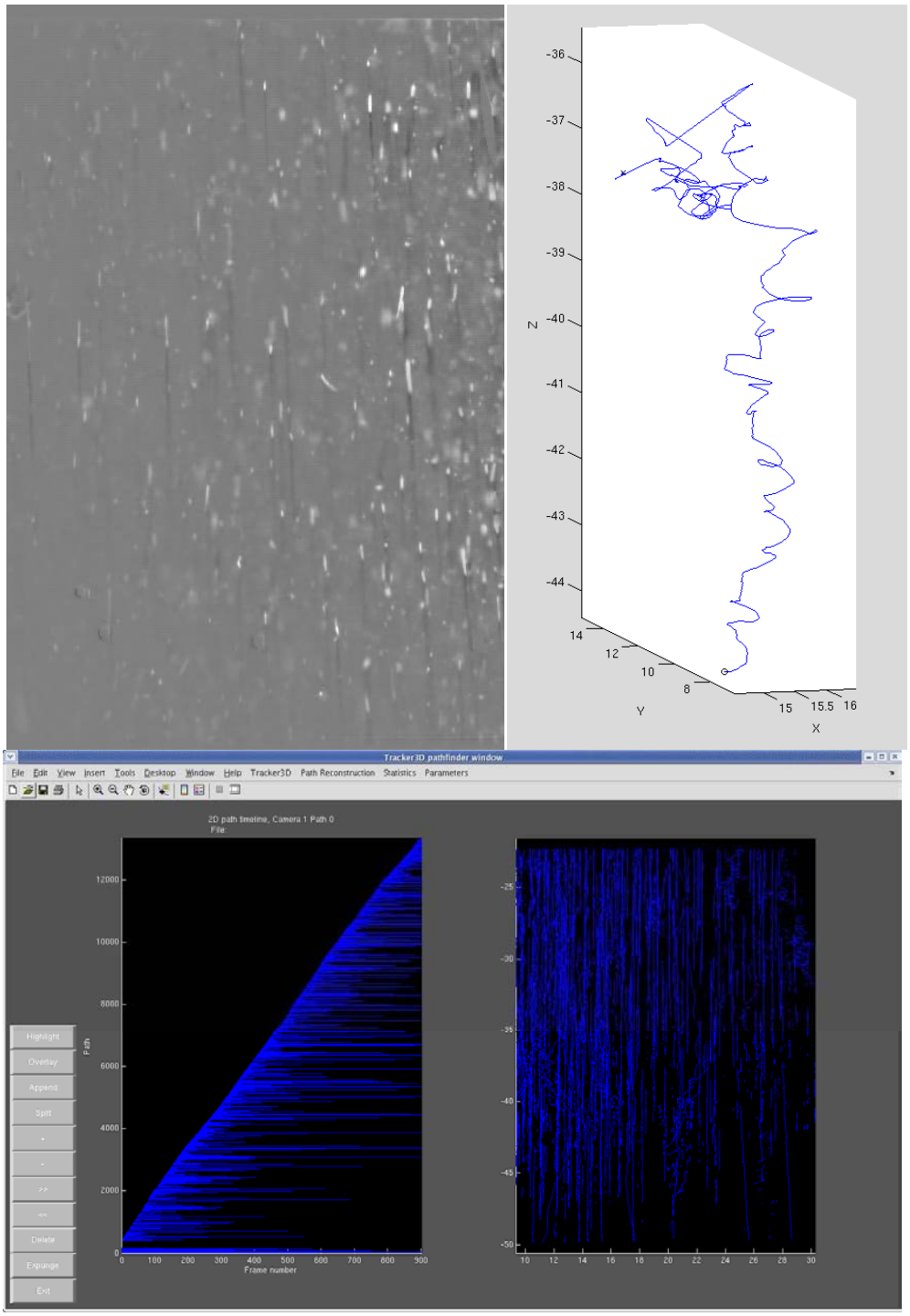

Figure 4. Swimming behavioral of protists at Station 26 in East Sound on May 4, 2006. Top left: A processed video frame from motion tracking of a natural plankton assemblage in a water sample collected at 7 m depth. The field of view is approximately 20 mm by 30 mm. Bottom: Screen capture of the tracking software, Tracker3D, which detected and tracked over 200 targets per video frame, including large numbers of chain diatoms and ciliates (see Figure 5). Top right: the 3-D trajectory followed by a large swimming ciliate over 23.3 s (probably Laboea strobilia). The straight segment at the top of the plot is a "hop" during which the cell moved approximately 2 mm in less than 100 ms.

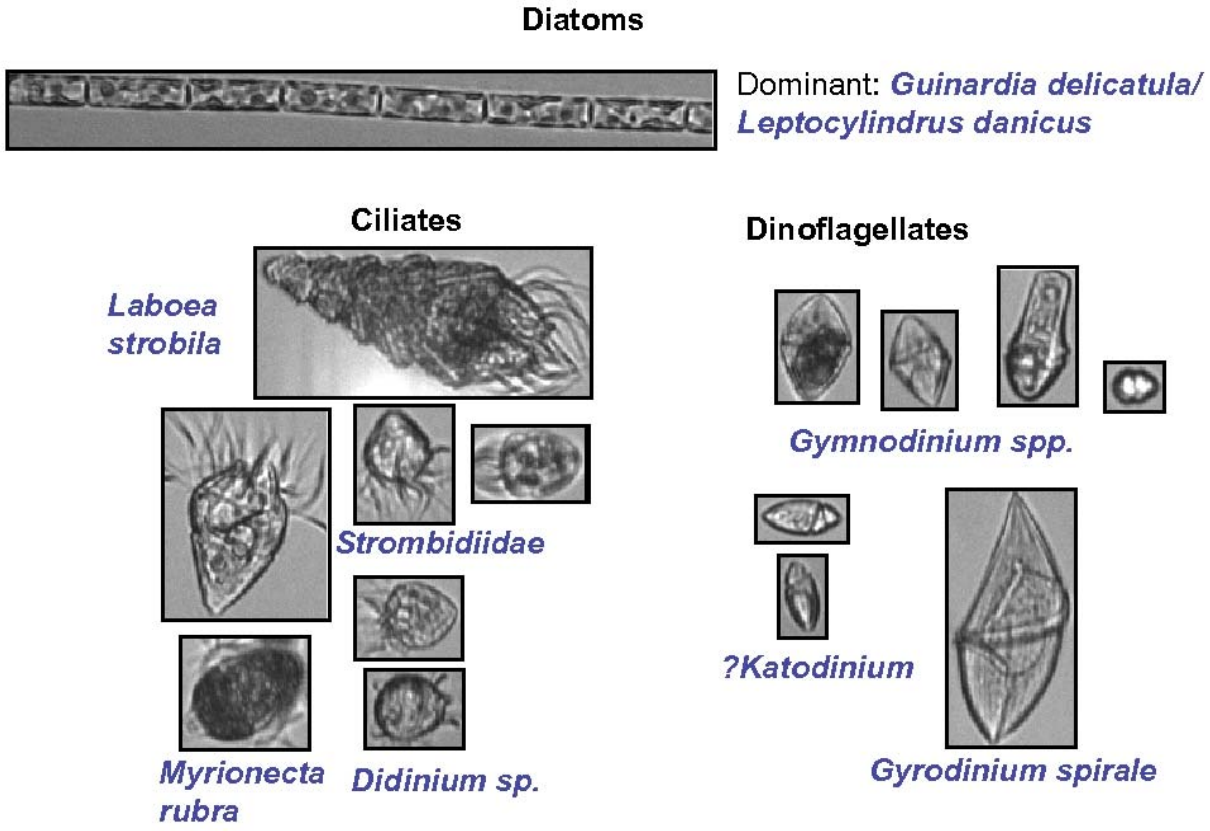

Figure 5. Dominant protistan taxa in the East Sound plankton community shown in Figure 4, indicated by simultaneous FlowCAM analysis of the same water sample.